

Fig.1A.

M V N R S V A F S A F V L I L F V L A I  
1 ATGGTGAATCGGTGGTTCGGTTCTCCGGTTCGTTCTGATCCTTTTCGTGCTCGCCATC  
S  
61 TCAGGTTATCAAATCCTTAGTTCATTTATTGAATATGATAGTATTATATCTTTATGG  
intron  
D I A S V S G E  
121 TTTTATGTGTTCTGACAAAGTTGCAAATATTGAGTAGATATCGCATCCGTTAGTGGAGAAC  
L C E K A S K T W S G N C G N T G H C D  
181 TATCGAGAAAGCTAGCAAGACATGTGCGGAAACTGTGGCAATACGGGACATTGTGACA  
NcoI  
N Q C K S W E G A A H G A C H V R N G K  
241 ACCAATGTAAATCATGGGAGGGTGGCCCATGGAGCGTGTGCATGTGCGTAACGGGAAAC  
HindIII  
H M C F C Y F N C K K A E K L A Q D K L  
301 ACATGTGTTTCTGTACTTCAATTGTAAAAAGCCGAAAGCTTGCTCAAGACAACTTA  
HindIII  
K A E Q L A Q D K L N A Q K L D R D A K  
361 AAGCCGAACAACCTCGCTCAAGACAACTTAATGCCCAAAAGCTTGACCGTGATGCCAAGA  
K V V P N V E H P  
421 AAGTGGTCCAAACGTTGAACATCCG



Fig.1B.

1 GTGCCCCGGGTCACGAAGTTCGGCACATCTTAGCGTTATGCATAAGTCAAAAATGGCCAA

M A K

61 AAATTCAGTTGCTTCTTTGCAATTGTGCCTGCTTCTTTTCATTCTTGCTATCTCAGAAAT

N S V A F F A L C L L F I L A I S E I

121 CAGATCGGTGAAGGGGAATTATGTGAGAAGGCAAGACATGGTCTGGAAATTGTGG

R S V K G E L C E K A S K T W S G N C G

181 CAATACAAGACACTGTGATGACCAGTGCAAGTCTTGGGAGGTGCAGCCCATGGAGCTTG

N T R H C D D Q C K S W E G A A H G A C

241 TCACGTGCGGGTGGAAACACATGTGCTTCTGCTACTTCAACTGTCCCAAAGCCCAGAA

H V R G G K H M C F C Y F N C P K A Q K

301 GTTGGCTGAGGATAAACTCAGAGCAGCAGAGCTAGCAAGGAGAAGATAATATTGGAGC

L A E D K L R A A E L A K E K N N I G A

361 TGAAGAAGTGCCCTTCAGCCACACCTTGAGTACTAACAAA

E K V P S A T P



Fig.2A.

M A K N S V A F L A F L L L L F V  
1 GGCACGAGTAATGGCCAAAATTACAGTTGCTTTCTTAGCATTTCTTCTGCTTCTTTTCGT  
L A I S E I G S V K G E L C E K A S K T  
61 TCTTGCTATCTCAGAAATCGGATCGGTGAAGGGGAATTATGTGAGAAGGCAAGCAAGAC  
W S G N C G N T R H C D D Q C K S W E G  
121 ATGGTCTGGAATTGTGGCAATACAAGACACTGTGATGACCAGTGCAAGTCTTGGGAGGG  
A A H G A C H V R G G K H M C F C Y F N  
181 CGAGCCCATGGAGCTTGTCACTGCGCGGTGGGAAACACACATGTGCTTTTGTACTTCAA  
C S K A Q K L A Q D K L K A D K L A K E  
241 CTGTTCCAAGCCCAAGAGCTGGCTCAGGATAAACTCAAAGCCGACAAGCTCGCCCAAGGA  
K S E A E K V P A T P  
301 GAAGAGTGAAGCCGAAAAGGTGCCAGCTACACCTTGAGTACTAAACAAGTGTGTATGATT  
361 ATGAATAAGAGAAAATGCTTTCTAGTTACCATATTTAGCATTTCTAATGTGTAATGTT  
421 TGTTGCTTTTGGAACTAAATTGCTTAACTATGATTCAGCTAATAATGTTTAAAGTATATA  
481 ATATAAGTTATCTTATTTTGAAGCCTGTAAAAAATAAAAAA



Fig.2B.

M A K N S V A F F A F V  
1 CGGCACGAGGCACAATCTCAAAAATGGCCAAAATTCGGTGTCTTCTTTCATTGTCC  
L L L F V L A I S E I G S V K G E L C E  
61 TGCCTCTTTCGTTCTTCTATCTCAGAAATTGGATCGGTGAAGGGAGAATTATGTGAGA  
K A S K T W S G N C G I T S H C D N Q C  
121 AGGCAAGCAAGACATGGTCTGGAAATTGTGGCATCACATCACACTGTGACAACCAAGTGCC  
R S W E G A I H G A C H V R G G K H M C  
181 GGTGCGGGAGGTGCAATCCATGGAGCTTGTACGTGCGCGGTGGGAAACACATGTGCT  
F C Y F N C S K A D E L A K E K I E A E  
241 TCTGCTACTTCAACTGTTCCAAAGCCGATGAGCTCGCGAAGGAGAAGATTGAAGCCGAAA  
K M P A T P  
301 AGATGCCAGCCACACCTTGAGTACTAACAATGCTATATGATTATAAAGAGAGAAAT  
361 GCTTCTAAAAAATAAAAAA

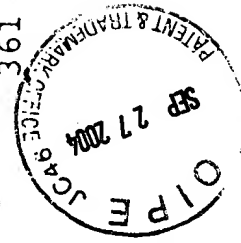
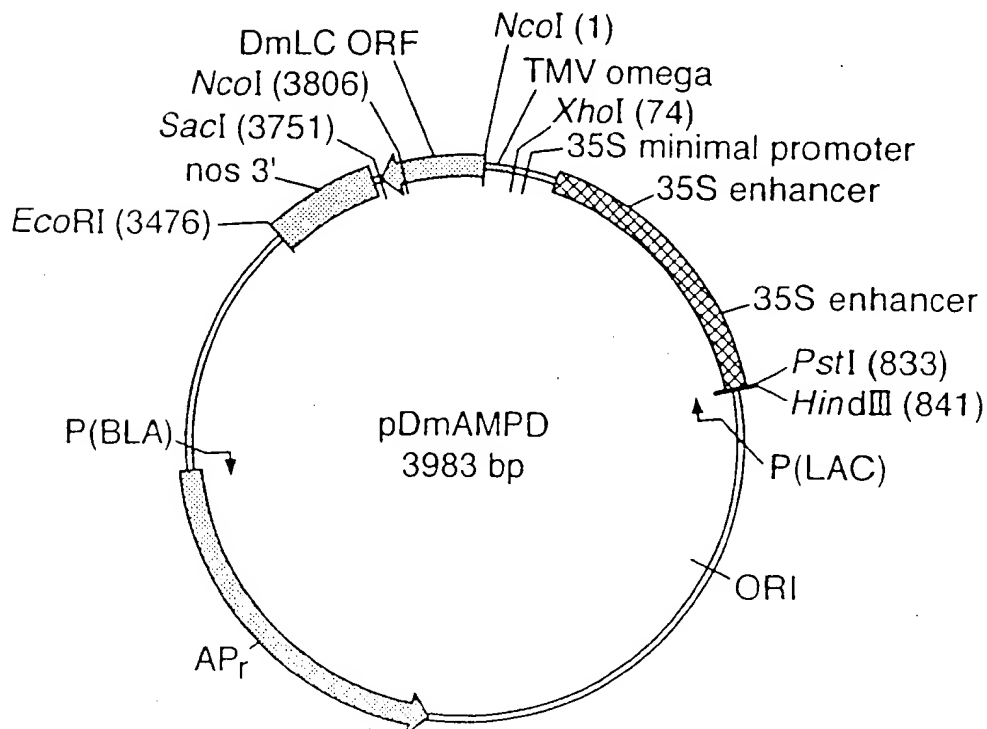
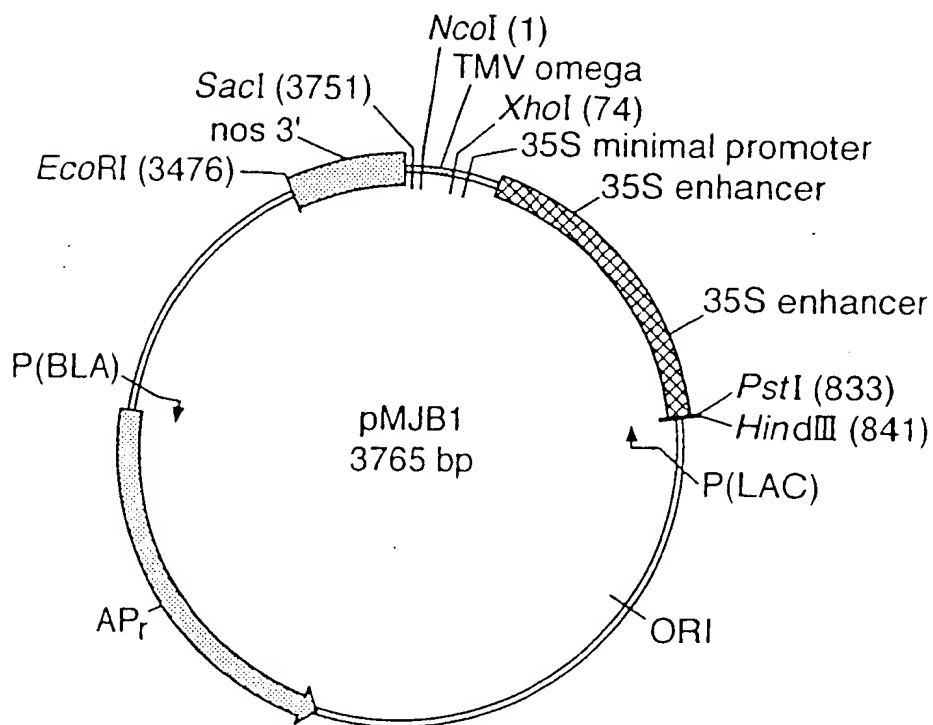


Fig.3.

M V N R S V A F S V F V L I  
1 GGCACGAGCCTATTAAAAAATGGTGAATCGATCGGTGCTTTCTCCGTGTTCTGAT  
L F V L A I S D I T S V R G E V C E K A  
61 CCTTTTCGTGCTCGCCATCTCAGATATCACAAAGTGTGAGAGGAGAAGTATGCGAGAAAGC  
S K T W S G N C G N T G H C D N Q C K Y  
121 TAGCAAGACATGGTCAGGAAACTGTGGCAACACGGGACACTGTGACAAACCAATGTAATA  
W E G A A H G A C H V R G G K H M C F C  
181 CTGGAGGGGGCCCATGGGGCGTCCACGTGCGTGGAGGGAACACATGTGTTCTG  
Y F K C P K A E K L A Q D K V N A Q E L  
241 CTAATTCAAGTGTCCTCCAAAGCCGAAAGCTTGCTCAAGACAAAGTTAATGCCCAAGAGCT  
D R D A K K V I P N V E H P  
301 TGACCGTGATGCCAAGAAAGTGATTCCCGAACGTTGAACATCCGTGAAAGGGTCGGTTTCT  
TAAATAGAAAGTCTTAGATTACGAATGCGAATAACTATAGAAAAATGTTGCTAAATGTC  
421 ACATTATAATTAGAACTTTATGATTGTTGTCAATAGGGCATTTTCTGTTAGTATGATGT  
481 GTAATAAGGTGATGCTTTTATGCTTTTCGTGCGTAAGAGTTTTCGACTATGTGTAATAAA  
541 GAAAGGTCTTTTTTTTTTAAAAAATAAAAAAAAAAAAAA



Fig.4.



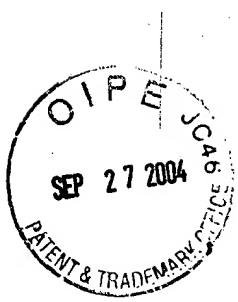


Fig.4 (Cont).

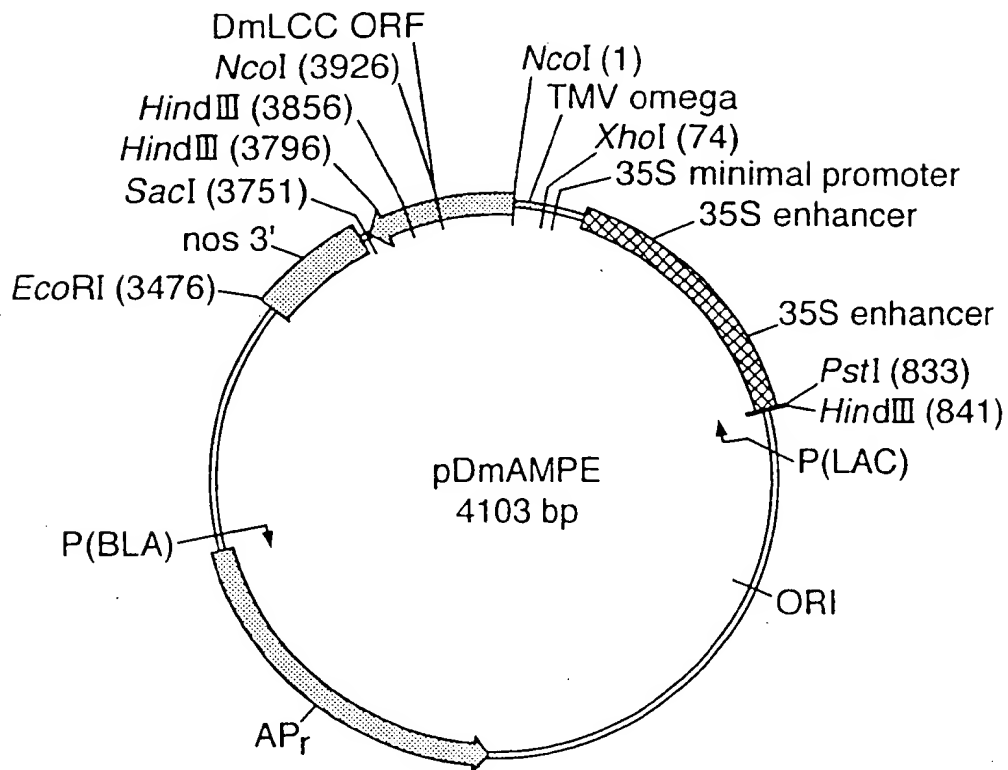
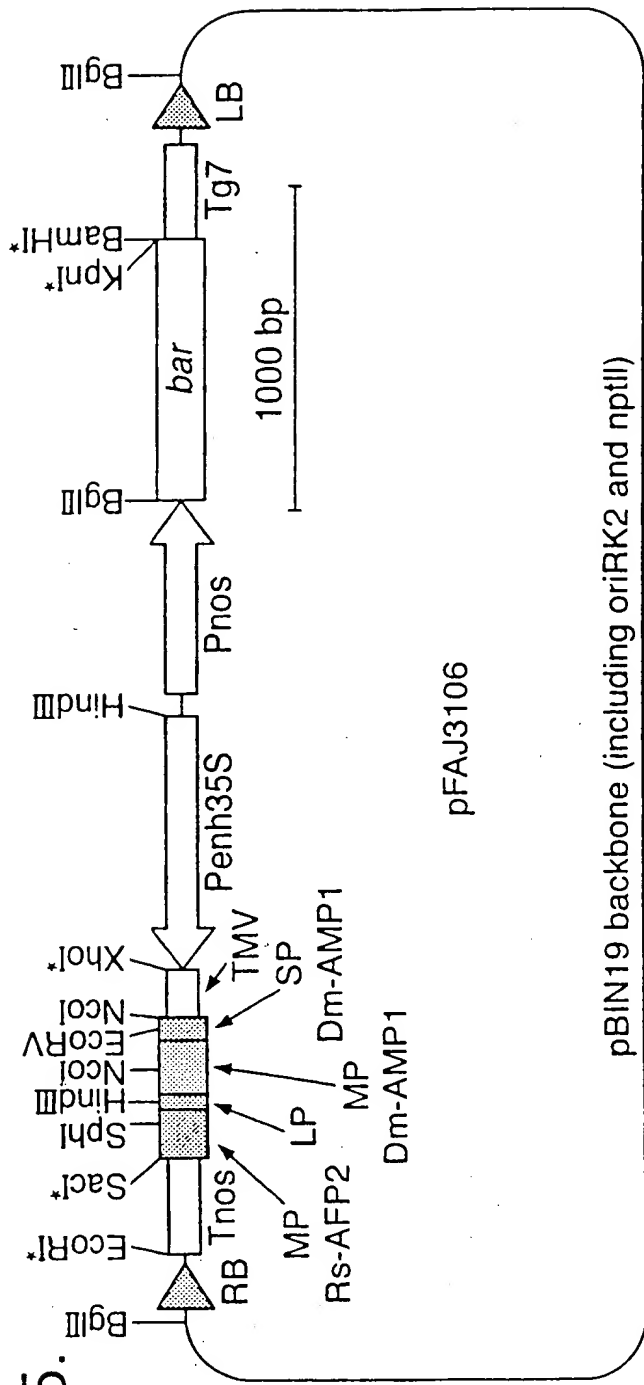


Fig.5.



#### Symbols

RB: right border of T-DNA

Tnos: terminator of T-DNA nopaline synthase gene

MP Rs-AFP2: mature protein domain of Rs-AFP2

LP: first 16 AA of Dm-AMP1 C-terminal propeptide and subtilisin-like protease recognition site IGKR

MP Dm-AMP1: mature protein domain of Dm-AMP1 cDNA

SP Dm-AMP1: signal peptide domain of Dm-AMP1 cDNA

TMV: tobacco mosaic virus 5' leader sequence

Penh35S: promoter of 35S RNA of cauliflower mosaic virus with duplicated enhancer region

Pnos: promoter of T-DNA nopaline synthase gene

bar: basta resistance encoding gene

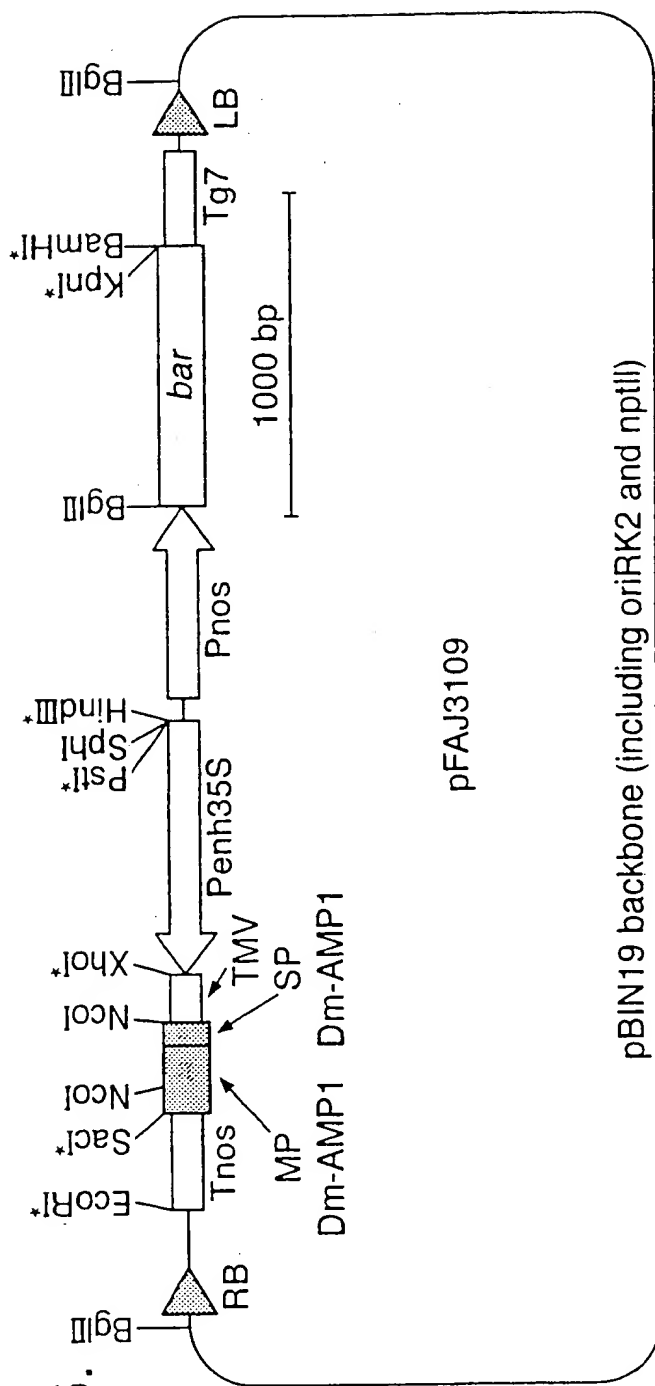
Tg7: terminator of T-DNA gene 7

LB: left border of T-DNA

\*: unique restriction site



Fig.6.



#### Symbols

RB: right border of T-DNA

Tnos: terminator of T-DNA nopaline synthase gene

MP Dm-AMP1: mature protein domain of Dm-AMP1

SP Dm-AMP1: signal peptide domain of Dm-AMP1 cDNA

TMV: tobacco mosaic virus 5' leader sequence

Penh35S: promotor of 35S RNA of cauliflower mosaic virus with duplicated enhancer region

Pnos: promotor of T-DNA nopaline synthase gene

bar: basta resistance encoding gene

Tg7: terminator of T-DNA gene 7

LB: left border of T-DNA

\*: unique restriction site

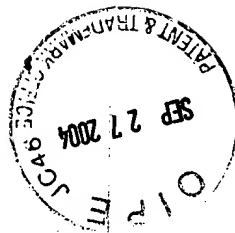


Fig.7. pFAJ3106

XhoI

CTCGAGTATTTTACAACAATTACCAACAACAACAACAACAACAATTACAATTACT

NcoI

ATTACAATTACACCATGGTGAATCGGTGGTGGTTCCTCCGGTTCGTTCGTGATCCTT

M V N R S V A F S A F V L I L

TTCGTGCTCGCCATCTCAGATATCGCATCCGTTAGTGGAGAACTATGCGAGAAAGCTAGC

F V L A I S D I A S V S G E L C E K A S

AAGACGTGTGGGCAACTGTGTGGCAACACGGGACATTTGTGACAACCAATGTAAATCATGG

K T W S G N C G N T G H C D N Q C K S W

GAGGTGCGGCCCATGGAGCGTGTTCATGTGCGTAACGGGAAACACATGTGTTTCTGTAC

E G A A H G A C H V R N G K H M C F C Y

TTCAATTGTAAAAAGCCGAAAGCTTGCTCAAGACAAAATTAAAGCCGAACACTCATC

F N C K K A E K L A Q D K L K A E Q L I

GGAAAGAGGCAGAGTTGTGCCAAAGCCCAAGTGGGACATGGTCAGGAGTCTGTGGAAC

G K R Q K L C Q R P S G T W S G V C G N

AATAACGCATGCAAGAATCAGTGCATTAGACTTGAGAAAGCACGACATGATCTTGCAAC

N N A C K N Q C I R L E K A R H G S C N

SacI

TATGTCTTCCAGCTCACAAGTGTATCTGTCTACTTTTCCTTGTTAATAGGAGCTC

Y V F P A H K C I C Y F P C - -



Fig.8.

pFAJ3109

XhoI

CTCGAGTATTTTACAACAATTACCAACAACAACAACAACAACAATTACAATTACT

NcoI

ATTTACAATTACACCATGGTGAATCGGTCGGTTGCGGTTCTCCGGCGTTCTGATCCTT

M V N R S V A F S A F V L I L

TTCGTGCTCGCCCATCTCAGATATCGCATCCGTTAGTGGAGAACTATGCGAGAAAGCTAGC

F V L A I S D I A S V S G E L C E K A S

AAGACGTGTGGGCAACTGTGGCAACACGGGACATTTGTGACAACCAATGTAAATCATGG

K T W S G N C G N T G H C D N Q C K S W

GAGGTGCGGCCCATGGAGCGTGTTCATGTGCGTAATGGGAAACACATGTGTTTCTGTAC

E G A A H G A C H V R N G K H M C F C Y

SacI

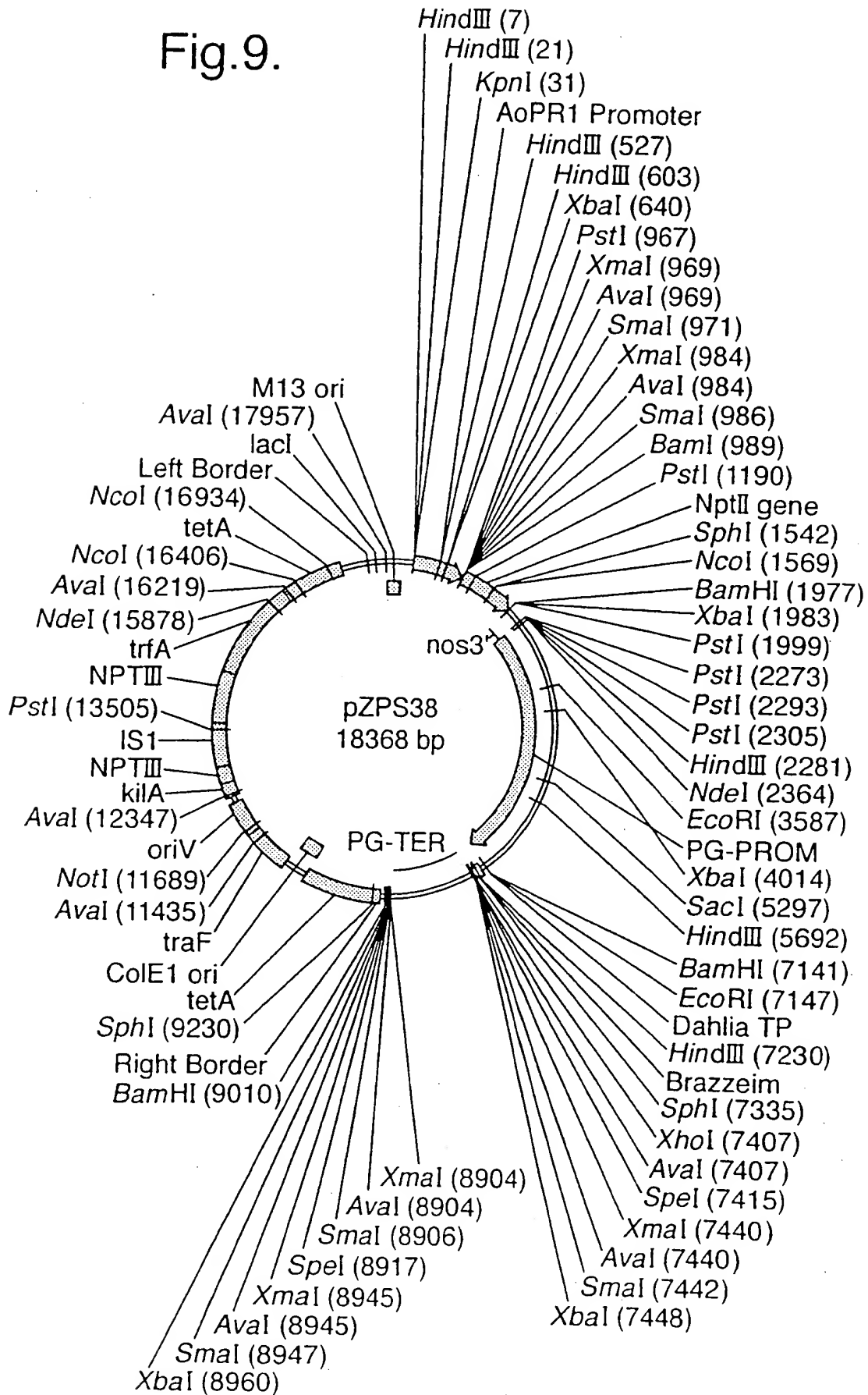
TTCAATTGTTGAGCTC

F N C -





Fig.9.



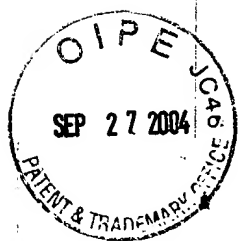


Fig.10.

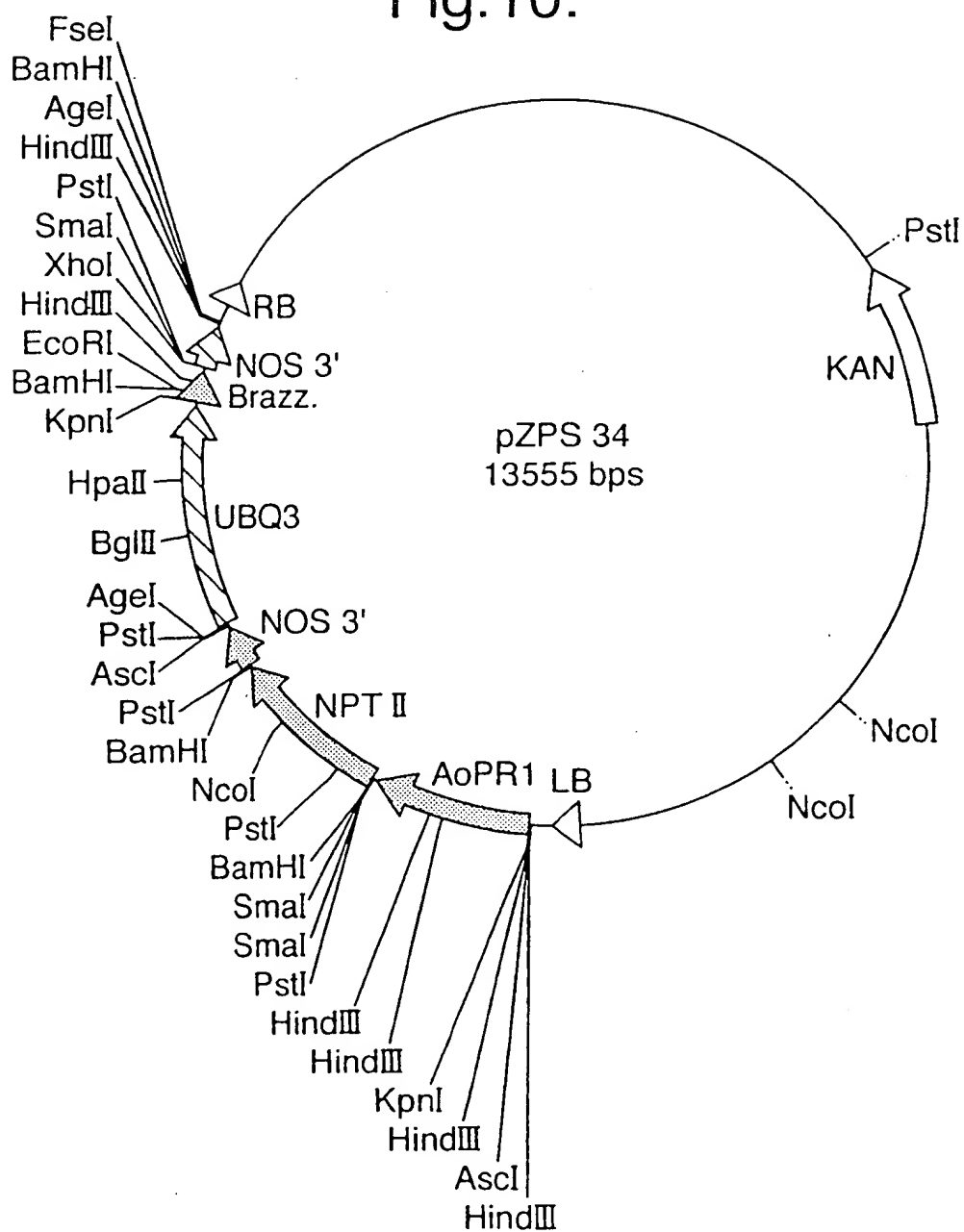




Fig.11.

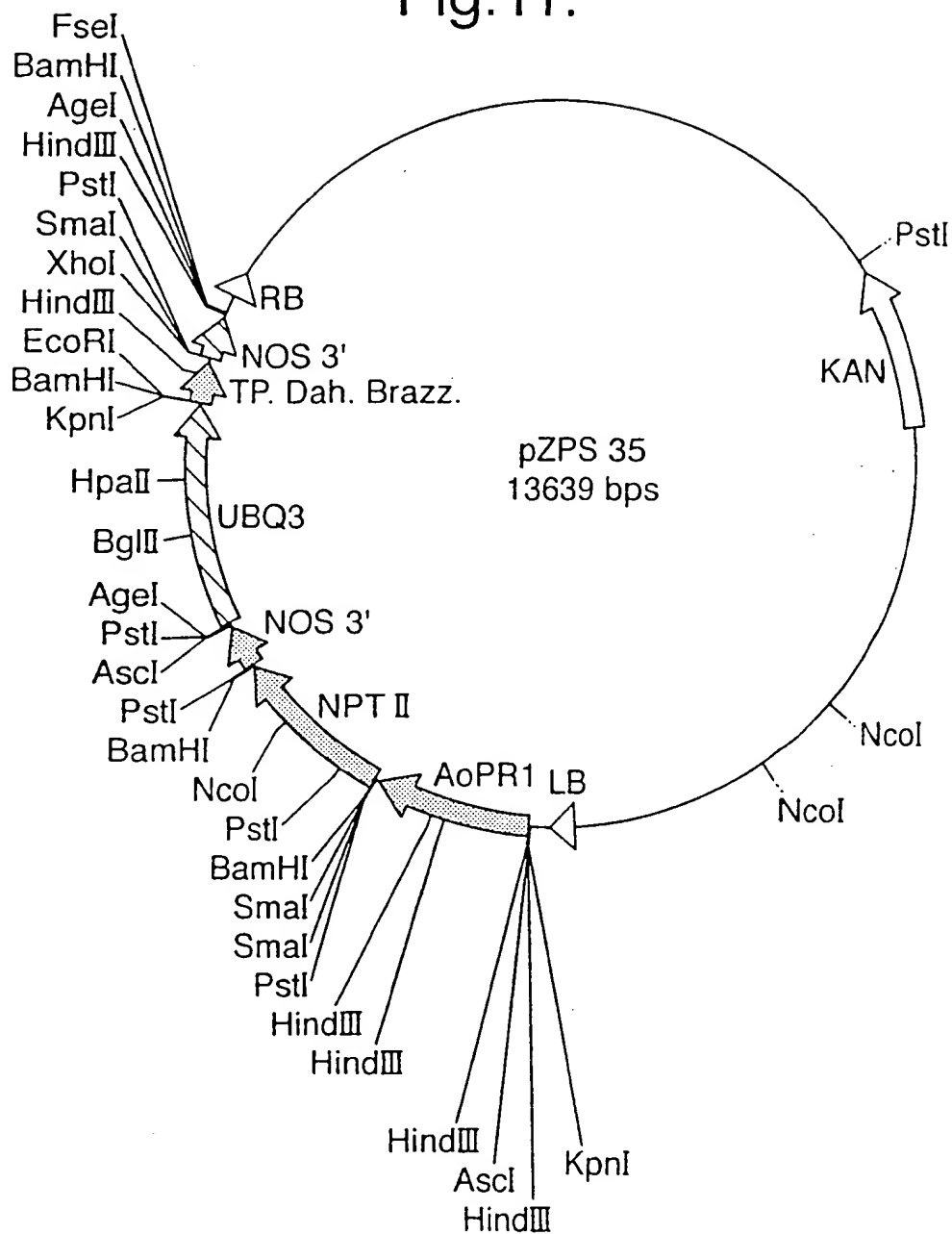
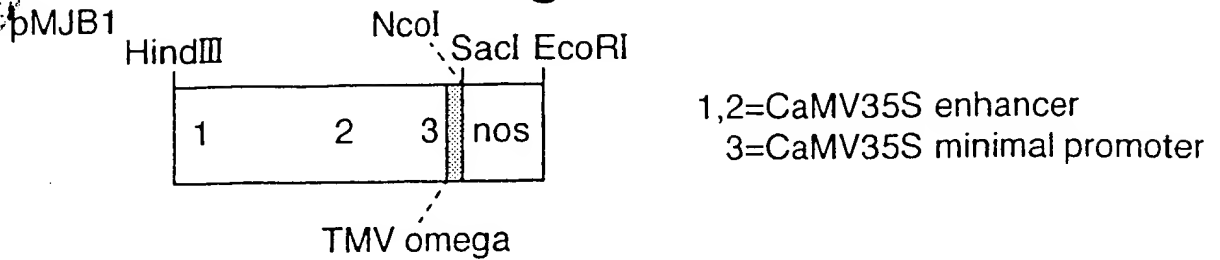


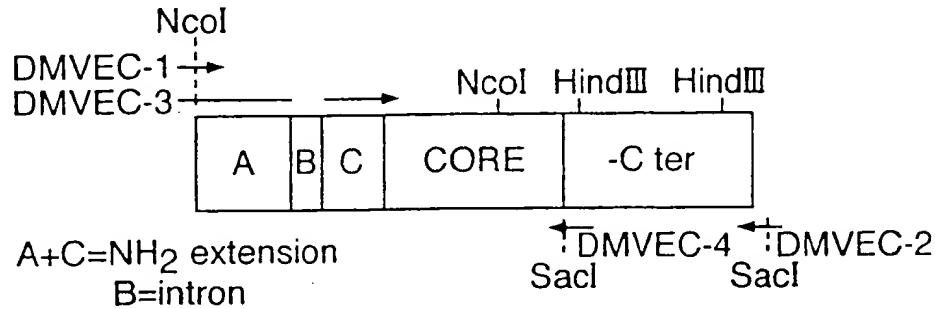




Fig.13.



Structure of DmAMP1 Gene and position of vector construction oligonucleotides

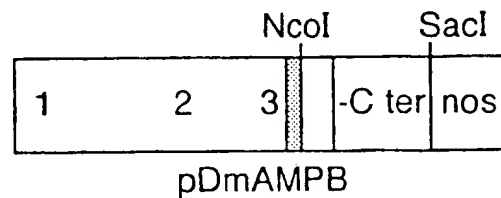
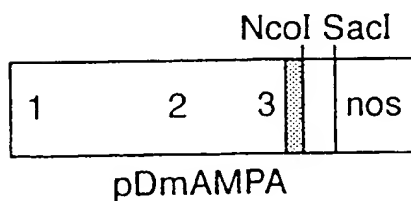


PCR Dahlia genomic DNA with DMVEC-1 and DMVEC-2, isolate 450 bp product.

PCR 450 bp DMVEC-1/DMVEC-2 PCR product with DMVEC 1 and 4.

Isolate 60 bp NcoI / SacI fragment, clone into pMJB1 NcoI / SacI=pDmAMPA.

Cut 450 bp DMVEC-1/DMVEC-2 PCR product NcoI / SacI . Isolate 180 bp NcoI / SacI fragment, clone into pMJB1 NcoI / SacI =pDmAMPB



PCR 450 bp DMVEC-1/DMVEC-2 PCR product with DMVEC 3 and 4.

Isolate 150 bp NcoI fragment, clone into pDmAMPA

and pDmAMPB NcoI=pDmAMPD and pDmAMPE

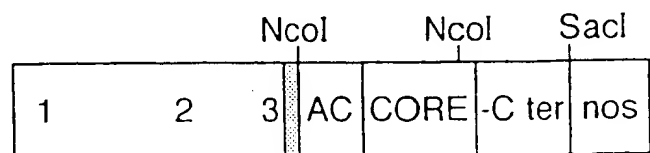
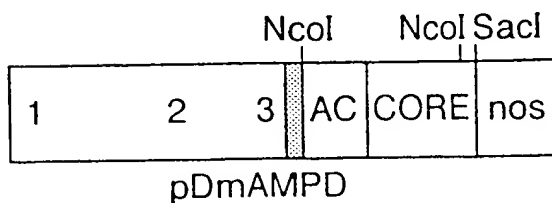


Fig.14.

Sequence  
ID No.6 Dm-AMP1

GAG CTT TGC GAG AAG GCT TCT AAG ACT TGG TCT GGA AAC

TGG GAG GGA GCT GCT CAT GGA GCT TGC CAT GTT AGA AAC

Sequence  
ID No.7 Dm-AMP2

GAG GTT TGC GAG AAG GCT TCT AAG ACT TGG TCT GGA AAC



Fig.14 (Cont).

TGC GGA AAC ACT GGA CAT TGC GAT AAC CAA TGC AAG TCT

GGA AAG CAT ATG TGC TTC TGC TAC TTC AAC TGC

TGC GGA AAC ACT GGA CAT TGC ... ..





Fig.15.

